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In any multicellular organism, organogenesis requires axial patterning to determine Antero-Posterior (AP), Dorsio-Ventral (DV), Proximo-Distal (PD) axes. Any deviation in these axes during development leads to congenital birth defects. In our model system, *Drosophila melanogaster* (a.k.a fruit fly), Dorsio-Ventral (DV) pattern marks first lineage restriction event. We have identified defective *protronicus* (*dve*-a Homeobox gene), an ortholog of SATB homeobox 1 (special AT-rich sequence binding protein 1), as a DV patterning gene. *dve* encodes a GATA-1 transcription factor, and is expressed in the downstream of *pannier* (*prn*-a GATA-4 transcription factor), and upstream of *wingless* (*wg*) in dorsal gene hierarchy. Loss-of-function of *dve* or *prn* results in dramatic dorsal eye enlargements, whereas gain-of-function suppresses the eye fate. We have demonstrated that *Wg* is a downstream target of Hippo growth regulatory pathway (highly conserved in eye). Furthermore, *Wingless* (*Wg*), which acts downstream of *dve*, also exhibits similar eye enlargement and suppression phenotypes and has been shown to play a role in growth. Here, we have shown that *dve* is a common DV patterning gene that is required to regulate the DV patterning of a common downstream target, *Wg* during growth and patterning of developing *Drosophila* eye. Our data (using Gain-of-function studies) states that activation of Hippo signaling in *dve*, *prn* expression domain results in change of head specific fate to an eye. We have tested retinal determination fate markers in these backgrounds. This study will address an important question, whether the axial patterning genes (*dve*, *prn*) and Hippo pathway regulates patterning and growth in other tissues and organs. The results from these studies will be presented.

Blocking Hippo Signaling suppresses the eye specific fate

Wild type Overgrowth Overgrowth

Hippo Signaling suppresses eye fate by ectopically inducing Wingless (Wg)

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Canton-S

A	B	C	D
<i>hpo^{-/-}</i>	<i>wts^{-/-}</i>	<i>yki^{-/-}</i>	
Dve Elav	Dve Elav	Dve Elav	Dve Elav

(I) In *hpo* and *wts* Loss-of-function clones, Dve expression domain increases

A'	B'	C'	D'
<i>hpo^{-/-}</i>	<i>wts^{-/-}</i>	<i>yki^{-/-}</i>	
Dve	Dve	Dve	Dve

(II) In *yki* Loss-of-function clones Dve expression domain decreases

A''	B''	C''	D''
<i>hpo^{-/-}</i>	<i>wts^{-/-}</i>	<i>yki^{-/-}</i>	
Elav	Elav	Elav	Elav

Hpo regulates Dve expression in the dorsal eye

[illegible]

Figure 1: Wg signaling is the downstream target of Hippo pathway in eye.

Schematic of Hippo Pathway: The diagram shows the regulatory network. At the top, *Ft* (Fat) and *D* (Disc-large) are shown. *Ft* inhibits *Yki* (Yorkie). *D* inhibits *Wts* (Warts). *Wts* inhibits *Yki*. *Yki* inhibits *Wg* (Wingless). *Wg* inhibits *Htn* (Hedgehog) and *Tsh* (Tissueless). *Htn* and *Tsh* inhibit *wg* (wingless). *wg* promotes *Morphogenetic Furrow (MF) progression*.

Gene Expression Diagram: A vertical diagram shows the expression of *hpo*, *wts*, *yki*, and *wg* in the eye. *hpo* is expressed in the dorsal eye (red arrow pointing down). *wts* is expressed in the ventral eye (green arrow pointing down). *yki* is expressed in the dorsal eye (red arrow pointing up). *wg* is expressed in the ventral eye (green arrow pointing up).

Microscopy Images: The images show the expression of *Wg* (green) and *Elav* (red) in the eye. The top row shows wild-type eyes (A) and *hpo>yki^{RNAi}* eyes (B). The bottom row shows *yki* expression in wild-type eyes (A') and *wg* expression in wild-type eyes (B').

Caption: Figure 1. Wg signaling is the downstream target of Hippo pathway in eye. **A**, **B**, *Wg* expression in the eye of wild-type and *hpo>yki^{RNAi}* flies. **A'**, **B'**, *yki* expression in the eye of wild-type and *wg* expression in the eye of wild-type flies. **Scale bar**, 100 μm.

expression domain

dve-Gal4GFP *dve>hpo* *dve>wts* *pnrGal4GFP* *pnr>hpo* *pnr>wts*

A B C D E F

A' GFP B' GFP C' GFP D' GFP E' GFP F' GFP

0 50 100 150 200 250 300

dve-GFP *dve-GFP>hpo* *dve-GFP>wts* *pnr-GFP* *pnr-GFP>hpo* *pnr-GFP>wts*

Inactivating Hippo Signaling increases *dve*- and *pnr*- expression domain

dve-Gal4GFP *dve>yki^{ts1A}* *pnrGal4GFP* *pnr>yki^{ts1A}*

A B C D

A' GFP B' GFP C' GFP D' GFP

0 50 100 150 200 250 300

dve-GFP *dve-GFP>yki^{ts1A}* *pnr-GFP* *pnr-GFP>yki^{ts1A}*

Sample size, N=5

changes eye specific fate to head and antennal fate

Based on –

- (i) Suppression of Eyes absent (Eya) and Dachshund (Dac)
- (ii) Upregulation of Wingless (Wg) and Homothorax (Hth)
- (iii) Change of eye specific fate to antenna (Ct)

- Components of Hippo signaling pathway interacts with members of DV patterning pathway.
- Both these pathways interact (Antagonistically) with each other.
- Cell Fate: Activating Hippo signaling suppresses *dve*, *pnr* expression, modulates wingless and changes head, antenna fate specific cells to an eye.
- Growth regulatory pathways regulates axial patterning genes and contributes to eye development

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